

Amino acids in nectar enhance longevity of female *Culex quinquefasciatus* mosquitoes

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ABSTRACT

Culex mosquitoes feed on a wide range of nectars consisting of mostly carbohydrates and amino acids, however, little is known about the utilization and effects of these different carbohydrates and their accompanying amino acids on longevity. *Culex quinquefasciatus* larvae were reared on low- and high-quantity food diets to produce adults that were nutritionally representative of wild-caught and laboratory-reared mosquitoes. Emerging adults reared on low- or high-quantity food diets as larvae were then provided *Lantana camara* nectar mimics containing mixtures of carbohydrates and amino acids to evaluate effects of nectar amino acids on longevity. Carbohydrates (with or without amino acids) were a critical component of the adult diet, and in their absence, adult mosquitoes died within 3–5 days. The nectar mimic that contained both carbohydrates and amino acids did not increase adult longevity of males originating from either poorly or well-fed larvae. However, females receiving adult diets containing both carbohydrates and amino acids lived 5% longer than females fed adult diets with only sugar.

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1. Introduction

The discovery that amino acids are the second most abundant component of nectars (Baker and Baker, 1973) and that some insects show a preference for carbohydrate sources containing amino acids (Alm et al., 1990; Mevi-Schutz and Erhardt, 2003b), has led to an increased interest in the role that they play in insect life histories (Baker and Baker, 1973). Some insects rely on nectar as a primary source for amino acids (Baker, 1977), but others feed on fruit, dung, pollen or even blood to obtain supplements (Baker and Baker, 1973; Foster, 1995). Feeding on pollen by female *Aedes aegypti* L. mosquitoes (Eischen and Foster, 1983) or ingestion of amino acids and carbohydrates by various species of male and female butterflies has been reported to enhance longevity, fecundity or both (O'Brien et al., 2003; Mevi-Schutz and Erhardt, 2005; Beck, 2007), although effects are typically not observed in laboratory animals reared from well-fed larvae (Bauerfeind and Fischer, 2005; Mevi-Schutz and Erhardt, 2003a; Hill and Pierce, 1989).

Certain flowers contain high levels of amino acids, and have been studied as potentially important sources for amino acids contributing to insect reproduction (Baker and Baker, 1973).

Common lantana (*Lantana camara* L. (Verbenaceae)) is an example of a butterfly flower that contains a high amino acid concentration (16 $\mu\text{mol/ml}$) as well as the sugars, fructose, glucose and sucrose (Alm et al., 1990). *Lantana camara* is a widely used ornamental and is naturalized in many temperate regions (Morton, 1994). Nectar mimics of *L. camara* have been used in numerous feeding preference studies with butterflies (Mevi-Schutz and Erhardt, 2003b; Alm et al., 1990) and are a known attractant for the tsetse fly (*Glossina* spp.) in Africa (Syed and Guerin, 2004).

The success of mosquitoes as disease vectors relies heavily on prolonged survival, to feed on multiple hosts and incubate pathogen development (Woodring et al., 1996). Feeding on nectar and honeydew enhances mosquito longevity and is also a source for flight energy (Nayar and Sauerman, 1971a, 1971b, 1975; Gary and Foster, 2004). When not feeding on nectar, anautogenous species feed on blood as a source of amino acids to make vitellogenin for egg production (O'Meara, 1987). Feeding pollen to anautogenous *Ae. aegypti* adults increased longevity, egg production in the lab and, if vertebrate hosts are unavailable, pollen feeding may enhance longevity and perhaps fecundity in field mosquitoes (Eischen and Foster, 1983). Adult *Culex nigripalpus* Theobald have been observed feeding on *Lantana* in the field (Haeger, unpublished data, cited in Nayar, 1982), but it remains unclear whether amino acids present in *Lantana* nectar will contribute to increased survival or fecundity. *Culex* mosquitoes are common throughout the United States, and *Culex quinquefasciatus*

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14. ABSTRACT Culex mosquitoes feed on a wide range of nectars consisting of mostly carbohydrates and amino acids however, little is known about the utilization and effects of these different carbohydrates and their accompanying amino acids on longevity. Culex quinquefasciatus larvae were reared on low- and highquantity food diets to produce adults that were nutritionally representative of wild-caught and laboratory-reared mosquitoes. Emerging adults reared on low- or high-quantity food diets as larvae were then provided Lantana camara nectar mimics containing mixtures of carbohydrates and amino acids to evaluate effects of nectar amino acids on longevity. Carbohydrates (with or without amino acids) were a critical component of the adult diet, and in their absence, adult mosquitoes died within 3?5 days. The nectar mimic that contained both carbohydrates and amino acids did not increase adult longevity of males originating from either poorly or well-fed larvae. However, females receiving adult diets containing both carbohydrates and amino acids lived 5% longer than females fed adult diets with only sugar.					
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Say are common nuisance species in the Southern U.S. *Culex* species are particularly important as vectors of St. Louis encephalitis virus (SLE) (Jones et al., 2002) and West Nile virus (WNV) (Sardelis et al., 2001; Molaei et al., 2007).

Many physiological attributes, such as body size and nutrient reserves, can affect the longevity of adult mosquitoes when reared under field conditions or in the laboratory. In the field, larvae are exposed to a wide range of nutritional conditions, and exposure to stress factors such as poor habitat quality or high competition, may result in reduced body size of adults (Day and Van Handel, 1986). Field-collected adult mosquitoes contain fewer nutritional reserves than those reared under controlled laboratory conditions (Day and Van Handel, 1986). Adult mosquitoes maintained in the laboratory solely on sucrose were able to store high levels of lipid and glycogen (O'Meara, 1987; Day and Van Handel, 1986), with levels typically greater than field caught individuals (Day and Van Handel, 1986). These data suggest that the condition of laboratory-reared mosquitoes may not necessarily reflect the nutritional state of mosquitoes in field settings.

Amino acids in nectar may provide an important metabolic substrate with the potential to enhance mosquito survival and vector capability. We evaluated whether amino acid supplementation in artificial nectar increased mosquito longevity. Effects of adult-acquired amino acids may only be apparent under nutritionally-stressed conditions. For example, in map butterflies, *Araschnia levana* L., the fecundity-enhancing effects of nectar amino acids were only detectable in adults that were poorly-fed as larvae (Mevi-Schutz and Erhardt, 2005) and preference for carbohydrates containing amino acids was only detected in adults that were poorly-fed as larvae (Mevi-Schutz and Erhardt, 2003b). Therefore, we reared mosquito larvae on high- or low-quantity food diets to mimic the range of nutritional conditions mosquitoes may encounter in the field. These high- and low-food larval treatments produced males and females that differed in body size and nutritional reserves at emergence. Nectar sugars are known to enhance longevity, so we assessed the effects of amino acids on adult longevity both in the presence of sugars and in the absence of sugars. Furthermore, because mating is known to enhance longevity in some mosquitoes (Liles and DeLong, 1960; Briegel and Kaiser, 1973), we tested whether larval nutrition had any effect on mating propensity.

2. Materials and methods

2.1. Mosquito rearing

A colony of *Cx. quinquefasciatus* established in Gainesville, FL in 1995 was used in this study. Larvae were reared in 2.5 L of water at a density of about 500/rearing container (35.5 cm × 48.3 cm × 6.4 cm) (28.0 ± 1.0 °C, 81.2 ± 0.1% RH, 14 L:10 D photoperiod). Diets were chosen to maximize differences in size without disrupting development time (Telang and Wells, 2004) (Table 1). Rearing larvae at a fixed density with varying amounts of food allowed us to evaluate effects of nutrition on larvae and subsequent effects on size and nutritional state of adults. The number of days from egg to pupation for the larvae reared on low-food diets was equal to that of the larvae reared on high-food diets.

2.2. Winglength and dry weight measurements

The effect of the larval rearing regimes on the size of resulting adults was initially determined by measuring winglength and dry weight. From each diet regime, samples of 10 newly emerged males and females were frozen (−20 °C). For each individual, wings were removed, mounted on glass slides, viewed at 10× magnifi-

Table 1

Feeding schedule of larval *Cx. quinquefasciatus* under low- or high-food diets. Larval food = 3% bovine liver powder (LP); 2% Brewer's yeast (BY) (30 g bovine liver powder and 20 g Brewer's yeast in 1 L of water), 2% hogchow (36 g finely ground hog chow in 1800 ml of water).

Days after hatch	Amount of food given per diet level (ml)	
	Low	High
1	50 ml; 3% LP; 2% BY	50 ml; 3% LP; 2% BY
3		50 ml; 2% hogchow
4	25 ml; 2% hogchow	50 ml; 2% hogchow
5		50 ml; 2% hogchow

cation with a dissection scope and photographed using a digital eyepiece camera with ScopePhoto 1.0 (Scopetek, Hangzhou). Measurements were made using a calibrated ocular micrometer from the alular notch to the distal end of the R₂ wingvein, fringe hairs excluded (SigmaScan Pro 5.0, SPSS, Inc., Chicago, IL) and average winglength was determined.

Dry weights were determined from the average of 5 replicate samples of 10 male and 10 female mosquitoes from each larval feeding regime. Samples were frozen at −20 °C just after emergence, frozen, freeze dried to constant mass and weighed.

2.3. Glycogen and lipid analyses

Initial nutritional reserves of the mosquitoes immediately after adult emergence were determined by measuring glycogen and triglyceride content. Samples that were previously freeze dried and weighed (see Section 2.2) were analyzed for glycogen (Van Handel, 1985) or sulphophovanillin assays for lipids (Van Handel, 1985, as modified by Hahn, 2005). In preparation for the assays, freeze-dried mosquitoes were homogenized in microcentrifuge tubes with 100 µl of saturated sodium sulfate, 200 µl of methanol, 100 µl of ultrapure water and 500 µl of 1:1 chloroform: methanol. Lipid solutions were washed through glass pipette columns of 0.2 g silicic acid with four rinses of 1 ml of chloroform to extract only neutral lipids (Hahn, 2005). Five replicate samples were completed for each sex and larval rearing regime.

2.4. Mating assay

Upon adult emergence, groups of 35 males and 35 females were placed together in separate 0.47 L paper containers (Solo Cup Company, Highland Park, IL), each representing one replicate. The mosquitoes were held for 15 days to ensure mating. The containers were modified with fabric screening to allow viewing through the top, and a 1.5 ml microcentrifuge tube was inserted into the side, to dispense treatment solutions via a piece of a saturated cotton dental wick (1 cm × 4.5 cm) (Richmond Dental, Charlotte, NC). This assay was conducted separately from the longevity assay (see Section 2.5), however, because the aim was simply to confirm that mating occurred and use that data to make inferences about the survival of mosquitoes in the longevity assay, the mosquitoes were maintained only on a 5% sucrose solution. Mosquitoes were maintained at 28.1 ± 1.0 °C, 81.2 ± 0.1% relative humidity and 14 L:10 D photoperiod. Five replicates were completed using mosquitoes fed a high-food diet as larvae and five replicates with those fed a low-food diet as larvae. Ten randomly chosen mosquitoes from each replicate were collected and kept at −20 °C until spermathecal dissections were performed. Upon dissection, the number of adult females from high-food larval diets or low-food larval diets containing sperm was counted to assess female mating.

2.5. Amino acid longevity assay

This experiment was set up similarly to the mating assay (see Section 2.4); however, mosquitoes were fed *ad libitum* on multiple treatments and the assay was conducted for the entire life of the mosquitoes. Treatments included a *L. camara* sugar mixture mimic (Alm et al., 1990; Mevi-Schutz and Erhardt, 2005), *L. camara* mimic with amino acids added, amino acids in water and a water only control. The nectar mimic contained 0.547 M sucrose, 0.282 M D-(+)-glucose, and 0.316 M D-(+)-fructose (Sigma, St. Louis, MO). The nectar mimic with amino acids additionally contained the nonessential amino acids L-alanine (0.718 mM), L-asparagine (0.421 mM), L-glutamic acid (0.326 mM), L-glutamine (0.931 mM), glycine (2.371 mM), L-proline (2.23 mM), and L-serine (1.37 mM) and the essential amino acids L-arginine (0.201 mM), L-threonine (0.672 mM), L-tyrosine (0.221 mM), and L-valine (0.137 mM) (Sigma, St. Louis, MO). Ten replicates of each treatment and five replicates of the water control were completed. The number of dead males and females were counted daily and the time to 50% mortality determined.

2.6. Statistical analyses

All statistical analyses were conducted using Minitab 15.1 (Minitab, Inc., State College, PA) or JMP (SAS Inc., Cary, NC). Assumptions of normality and homogeneity of variance were confirmed using graphical representations of the data and Levene's test ($P < 0.05$).

One-way ANOVAs were used to determine differences in winglength and dry weight among females or males from each larval rearing regime. Mean triglyceride and glycogen weights were obtained for each replicate group of 10 males and 10 females. Glycogen and lipid weights were divided by dry weight to correct for mass differences. Data for glycogen analyses were Ln transformed as necessary. A one-way ANOVA was used to determine differences in the mean corrected lipid or glycogen weights, and Tukey's HSD ($P < 0.05$) was used to assess differences between the means.

Male mosquitoes are known to have a significantly shorter lifespan than females of the same species (Liles and DeLong, 1960); therefore number of days to 50% mortality for each replicate was determined for males and females separately. Previous studies have also determined that a water diet supports life for only a few days compared to weeks on a sugar source (Nayar, 1986; Nayar and Sauerman, 1971a, 1971b, 1975), therefore treatments without sugar (water only and water + amino acids) and treatments with sugar (*L. camara* nectar mimic and *L. camara* nectar mimic + amino acids) were also considered separately. Data were used to build four multivariable ANOVA models containing all explanatory variables (adult diet, larval diet and their interaction). Tukey's HSD

($P < 0.05$) or two-sample *t*-tests ($P < 0.05$) were used to separate the means.

3. Results

3.1. Effect of larval nutrition on adult size, nutrient reserves and mating

Both males and females had longer wings and were heavier when reared on high-food larval diets (2.70 ± 0.05 (♂) or 3.49 ± 0.18 (♀) mm and 5.07 ± 0.16 (♂) or 9.11 ± 0.10 (♀) mg, respectively) than on low-food larval diets (2.41 ± 0.04 (♂) or 2.64 ± 0.06 (♀) mm and 2.70 ± 0.05 (♂) and 3.49 ± 0.18 (♀) mg, respectively) ($t = 6.90$, $df = 15$, $P < 0.001$ (♂); $t = 8.37$, $df = 12$, $P < 0.001$ (♀); $t = 14.02$, $df = 4$, $P < 0.001$ (♂); $t = 27.77$, $df = 6$, $P < 0.001$ (♀), respectively). Males and females originating from a high-food larval diet were 1.9 and 2.6 times heavier, respectively, than their low-food diet counterparts.

Both males and females had larger glycogen and lipid stores when fed a high-food diet as larvae (82.61 ± 9.70 (♂) or 96.44 ± 5.12 (♀) µg/mg glycogen and 113.31 ± 1.43 (♂) or 108.37 ± 2.41 (♀) µg/mg lipids, respectively) than when fed a low-food diet as larvae (9.38 ± 1.67 (♂) or 7.15 ± 0.56 (♀) µg/mg glycogen and 42.64 ± 2.83 (♂) or 33.84 ± 3.71 (♀) µg/mg lipids, respectively) ($t = 6.35$, $df = 4$, $P = 0.003$ (♂); $t = 16.73$, $df = 4$, $P < 0.001$ (♀); $t = 20.64$, $df = 5$, $P < 0.001$ (♂); $t = 26.24$, $df = 7$, $P < 0.001$ (♀), respectively). Males and females originating from a high-food larval diet stored nearly 9 or 13 times more glycogen and 4 or 3 times more lipids, respectively, than when fed a low-food diet as larvae. Larval diet had no effect on mating, 98% mated on high-food and 96% mated on low-food diets.

3.2. Survival analyses

3.2.1. Mosquitoes fed treatments without sugar

Adult diets lacking sugar (water only or water + amino acids) did not sufficiently support survival, with all individuals living <5 days (Table 2). Adding amino acids to water did not significantly increase longevity of males or females (Tables 2 and 3). Larval diet had the greatest effect on survival; males from a high-food larval diet lived twice as long as males fed a low-food larval diet and high-food larval diet females lived 50% longer than those reared on a low-food larval diet (Tables 2 and 3).

3.2.2. Mosquitoes fed treatments with sugar

Adding amino acids to the adult diet of females resulted in a 5% increase in survival of females overall, no matter what their larval diet (Fig. 1d and Tables 2 and 3); however, a benefit to survival was not observed in males when fed a similar diet (Fig. 1c and Table 2). As expected, adults fed treatments containing sugar lived at least

Table 2

Days to 50% mortality (LS means \pm SE) of male and female *Cx. quinquefasciatus* fed low- or high-food diets as larvae. Adult diets included no sugar treatments (water only, water + amino acids) and sugar treatments (*Lantana camara* nectar mimic and *L. camara* mimic + amino acids).

Larval diet	Adult diet	Days to 50% mortality (\pm SE) (LS means) ^a					
		♂	N	No. of mosquitoes	♀	N	No. of mosquitoes
Low	Water only	2.20 \pm 0.14	5	175	3.00 \pm 0.12	5	175
	Water + amino acids	2.33 \pm 0.13	6	210	3.00 \pm 0.11	6	210
Low	<i>L. camara</i> nectar mimic	30.00 \pm 1.04	7	245	32.71 \pm 0.98	7	245
	<i>L. camara</i> mimic + amino acids	32.00 \pm 1.12	8	280	35.25 \pm 0.92	8	280
High	Water only	5.00 \pm 0.10	5	175	4.80 \pm 0.12	5	175
	Water + amino acids	5.00 \pm 0.14	10	350	4.90 \pm 0.09	10	350
High	<i>L. camara</i> nectar mimic	27.33 \pm 0.99	10	350	36.70 \pm 0.082	10	350
	<i>L. camara</i> mimic + amino acids	27.60 \pm 0.94	10	350	38.00 \pm 0.082	10	350

^a All LS means derived from full models (adult diet, larval diet and their interaction).

Table 3

Results of a reduced model ANOVA on the effect of larval diet and adult diet on survival of male or female *Cx. quinquefasciatus*. Non-significant interaction terms were removed from the model.

Adult diet	Sex	Source	df	F	P
No sugar	Males	Whole model	2	251.43	<0.001*
		Larval diet	1	492.37	<0.001*
		Adult amino acids	1	0.23	0.634
		Error	23		
		Total	25		
No sugar	Females	Whole model	2	148.40	<0.001*
		Larval diet	1	289.98	<0.001*
		Adult amino acids	1	0.25	0.625
		Error	23		
		Total	25		
Sugar	Males	Whole model	2	5.91	0.007*
		Larval diet	1	11.34	0.002*
		Adult amino acids	1	0.51	0.480
		Error	31		
		Total	33		
Sugar	Females	Whole model	2	9.16	<0.001*
		Larval diet	1	14.43	<0.001*
		Adult amino acids	1	4.40	0.044*
		Error	32		
		Total	34		

Bold text indicates significant effect of our primary hypothesis.

* A significant effect ($P < 0.05$).

5 times longer than adults not fed sugar (Table 2). Interestingly, adult males fed sugar lived longer when fed a low-food diet as larvae (Fig. 1a and Tables 2 and 3), but adult females fed sugar lived longer when fed a high-food larval diet (Fig. 1b).

4. Discussion

Nectar mimics containing amino acids enhanced longevity in females from both high- and low-food larval rearing regimes, but had no effect on males. As expected, adults that were fed treatments containing sugar lived substantially longer than those that were denied a sugar source as adults. Our study also confirmed that larval diet played an important role in nutritional reserves and size on emergence, which in turn, affected adult survival. As expected, large, well-fed adult females survived longer as adults, but interestingly, smaller, poorly-fed males survived longer than large, well-fed males as adults.

Although the increase in survival was small for adult females fed amino acids as adults (~5%), it was statistically detectable and is concordant with results from a study focusing on *Ae. aegypti* fed pollen in their adult diet (Eischen and Foster, 1983). *Araschnia levana*, a butterfly fed amino acids in their adult diets (Mevi-Schutz and Erhardt, 2005) lived longer when fed amino acids as well, but only when fed poorly as larvae, which did not seem to matter for female *Cx. quinquefasciatus* in our study. Studies with butterflies have demonstrated a preference for adult diets that contain amino acids (Alm et al., 1990), particularly when nutritional deprivation occurred as larvae (Mevi-Schutz and Erhardt, 2003b). Effects of adult amino acid supplementation on performance may be difficult to detect when using laboratory-reared insects because they contain sufficient nutritional reserves for survival acquired from the larval stage. The use of laboratory-reared insects without taking into account larval nutrition may explain conflicting results of the effect of amino acids on the fecundity and longevity of various insects (Mevi-Schutz and Erhardt, 2003a, 2005; Eischen and Foster, 1983) and even among species living under similar

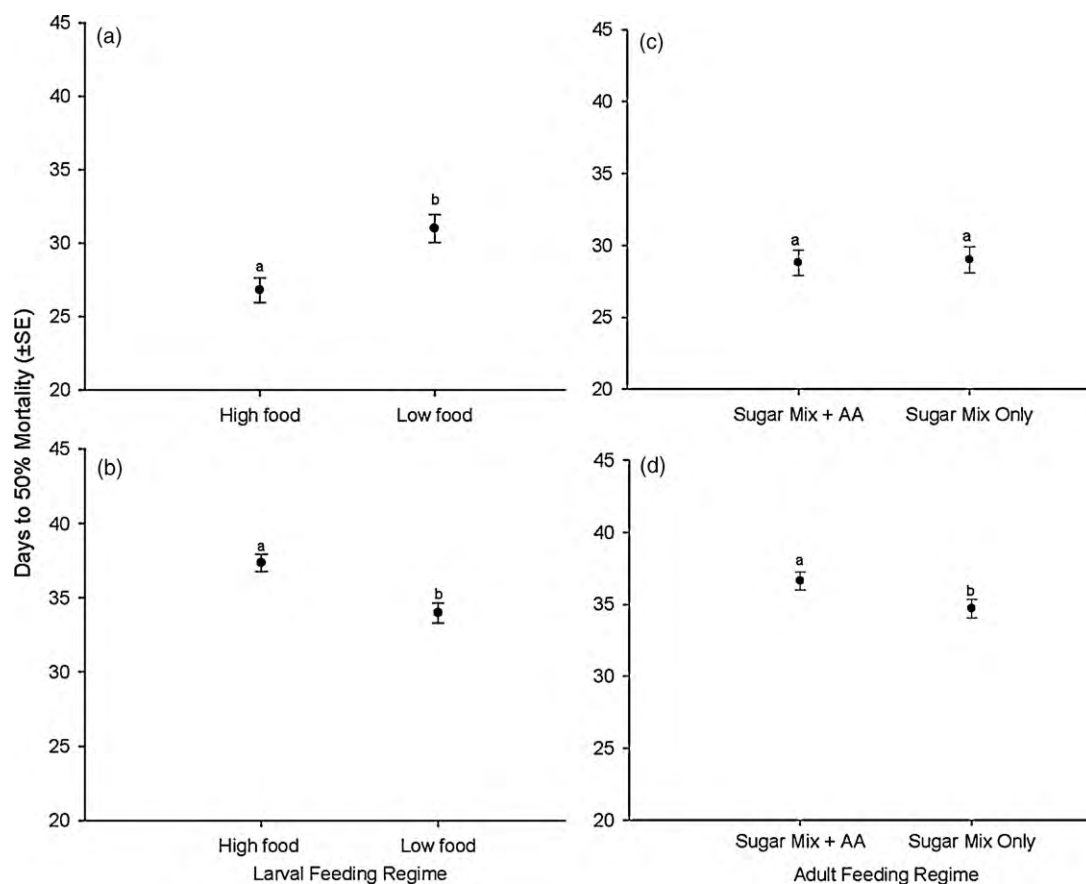


Fig. 1. Effect of larval diet on days to 50% mortality (LS means) of males (a) and females (b) and adult diet on males (c) and females (d) fed treatments with sugar. Treatments with similar letters are not significantly different (t -tests or Tukey's HSD, $P < 0.05$).

adult conditions (Beck, 2007). Additionally, the role of amino acids in nectar may differ between mosquito species; for instance, in autogenous mosquito *Aedes atropalpus* Coquillett whose larval proteins are directed toward vitellogenin in early adult life, blood feeding is unnecessary for developing an initial egg batch (Wheeler and Buck, 1996). In contrast, anautogenous mosquitoes such as some *Culex* spp. require amino acids from a blood meal as adults to stimulate vitellogenin production (Clements, 1956). Ingestion of amino acids, however, does not appear to have universally enhancing effects. Beck (2007) reported that longevity was increased only males of some species of butterflies, whereas other authors did not find a link between ingestion of amino acids and increased performance (Liles and DeLong, 1960; Mevi-Schutz and Erhardt, 2003a; Molleman et al., 2008; Bauerfeind and Fischer, 2009). However, our results suggest that the presence of amino acids in the adult diet may play a key role in enhancing survival of female mosquitoes.

Larval insects are exposed to many challenges in nature, including overcrowding and inadequate nutrition, with larval nutritional deprivation common in mosquitoes (Day and Van Handel, 1986). There was a substantial effect of larval diet on adult of both sexes in this study. Both male and female *Cx. quinquefasciatus* emerged with larger winglengths, higher dry weights and greater glycogen and lipid reserves when reared on a high-food diet as larvae compared to a low-food diet. In studies using laboratory-reared mosquitoes, females exposed to high-food resources likely have greater glycogen and lipid reserves and exhibit increased survival, and most closely resemble those females fed high-food diets in this study. In contrast to females, sugar-fed males in our study lived significantly longer when fed a low-food diet as larvae. This could be explained by reports that the association of males and females enhances female longevity and decreases survival of males (Liles and DeLong, 1960). Nearly all of the females in this study were mated, which may play a role in their increased longevity. However, based on their decreased longevity, males reared on a high-food diet as larvae may have suffered disproportionately from their association with females. Ng'habi et al. (2005) reported that male mosquitoes fed high-food diets as larvae were larger upon adult emergence and were more likely to be better competitors and mate earlier than smaller adult males that experienced nutritional stress as larvae. Increased mating occurrences may reduce survival similar to that reported for male *Saltella sphondylii* (Diptera: Sepsidae) (Martin and Hosken, 2004). This may, in part, explain why male *Cx. quinquefasciatus* that were reared on a high-food larval diet, and consequently had higher levels of lipids and glycogen upon emergence, would be potentially more sexually competitive and, in turn, would not survive as long as adults fed low-larval food diets.

Larval and adult nutrition critically affect not only survival, but also flight potential (Nayar and Sauerma, 1971a, 1971b, 1975), biting persistence (Nasci, 1991), and thus, disease transmission by mosquitoes (Nayar and Sauerma, 1975; Foster, 1995). The long amplification periods of arboviruses (>12 days) transmitted by *Culex* mosquitoes require a long lifespan (Turell et al., 2005), and mosquito survival is highly dependent on the timely ability to locate and feed on nectar, honeydew or other sources of carbohydrates and amino acids. Our findings that amino acids added to an adult diet enhance the survival of females no matter their larval diet may contribute to their potential for becoming successful disease vectors. Adults that encountered poor larval conditions emerge with low teneral reserves and without an immediate nectar and amino acid source will not have energy for flight or host-seeking. Some mosquitoes will not begin host-seeking until they have obtained an initial sugar meal (Foster, 1995) and adults with low nutritional reserves have been found to

be better vectors of WNV due to their reduced ability to avoid infection compared to adults with high nutritional reserves (Vaidyanathan et al., 2008). Feeding frequently on nectar to increase nutritional reserves is necessary for mosquitoes (Bidlingmayer and Hem, 1973; Foster, 1995) and because amino acids are abundant in these nectar sources (Baker and Baker, 1973), they may play additional roles in the diet of females. Future research may examine the effect of larval nutrition on the establishment and transmission of pathogenic agents, and the effects of adult diets rich in amino acids on the vector potential of mosquitoes.

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